

## Role of Mannose-binding Lectin-2 Promoter Genetic Variants in Susceptibility to Sepsis: A Meta and Trial Sequential Analysis

Hongyi Shao, Jianfeng Zhang, Dayong Wu, Xiaoyang Zhang, and Chunqiong Hu

Department of Emergency Intensive Care Medicine, The Central Hospital Affiliated to Shaoxing University, Shaoxing, Zhejiang, China

Received: 4 June 2025; Received in revised form: 2 August 2025; Accepted: 4 September 2025

### ABSTRACT

Mannose-binding lectin (MBL) is a critical component of the innate immune system, serving a vital role in the body's initial defense against pathogens. Sepsis, a severe condition triggered by an excessive immune response to infection, has been linked to variations in the MBL2 gene that affect MBL levels and functionality. Numerous studies across various populations have examined the role of MBL-2 promoter polymorphisms (H>L and Y>X), but their results have been conflicting. This study aims to investigate the genetic connection between MBL promoter polymorphisms and susceptibility to sepsis through a meta-analysis of previously published articles.

A thorough literature search was conducted using PubMed, Scopus, and ScienceDirect to locate relevant articles for the meta-analysis. Rigorous inclusion and exclusion criteria were implemented to ensure data accuracy. All analyses were performed using Comprehensive Meta-Analysis Software v4.

Seven studies were included, examining the role of MBL-2 promoter genetic variants in sepsis (H>L: n=3, sepsis cases: 449, control: 687; Y>X: n=6, sepsis cases: 1211, control: 1694). Egger's regression analysis and funnel plots suggested no publication bias. Heterogeneity analysis indicated homogeneity among the data. The meta-analysis showed no association between MBL-2 promoter variants and susceptibility to sepsis. The trial sequential analysis highlighted the need for further studies on MBL-2 promoter variants in sepsis to draw a definitive conclusion.

The promoter variants of the MBL-2 gene (H>L and Y>X) do not appear to increase the risk of sepsis. Further investigation is needed to confirm this conclusion, including more participants from diverse populations and larger sample sizes.

**Keywords:** Genetic predisposition to disease; Innate immunity; Mannose-binding lectin; Meta-analysis; Polymorphism; Sepsis

### INTRODUCTION

Sepsis is a severe condition caused by the body's

extreme response to an infection, which leads to widespread inflammation and potential organ failure.<sup>1</sup> In the year 2020, approximately 48 900 000 cases of sepsis were reported globally, resulting in the death of 11 000 000 individuals.<sup>2</sup> Notably, half of these cases occurred in individuals under five. Approximately 1.5% of sepsis cases that arise in hospitals have been reported in patients who were initially admitted for the treatment

---

**Corresponding Author:** Chunqiong Hu, B.Sc;  
Department of Emergency Intensive Care Medicine, The Central Hospital Affiliated to Shaoxing University Shaoxing, Zhejiang, China. Tel: (+86 137) 3539 1393, Fax: (+86 0575) 8558 0715, Email: huchunqiong\_214@sina.com

of other medical conditions.<sup>3</sup> These cases are associated with high mortality rates and longer hospital stays. The incidence and mortality rates for sepsis remain higher in lower-middle-income countries.<sup>2</sup> Treating a sepsis patient in higher-income countries typically incurs an average cost of \$32 000.<sup>4</sup> It is important to note that not all instances of infection lead to sepsis. However, a specific subset of individuals who are infected are at risk for developing sepsis. Environmental factors, host immunity, and genetics significantly influence an individual's susceptibility to sepsis.<sup>5,6</sup>

Innate immunity is integral to the immediate defense mechanism against invading pathogens by recognizing specific conserved molecular patterns. Upon encountering foreign pathogens, innate immune cells initiate complex pathways of immune responses, ultimately eliminating the pathogens and restoring homeostasis.<sup>7</sup> However, in sepsis, the host immune response becomes excessively vigorous, resulting in the release of uncontrolled proinflammatory cytokines, which leads to tissue injury and organ failure.<sup>8</sup> Concurrently, functional impairment of the innate immune system results in cell depletion, leading to improper pathogen clearance, predisposing patients to immunosuppression and secondary infections.<sup>9</sup> The balance between hyperinflammation and immune paralysis of the innate immune system is a major factor contributing to the complexity of sepsis and presents significant challenges for its therapeutic management.<sup>10</sup>

Mannose-binding lectin (MBL) is a vital protein that functions in the innate immune system, playing a crucial role in safeguarding the body against infections.<sup>11</sup> This protein recognizes and binds to specific carbohydrate patterns, including mannose and N-acetylglucosamine, which are found on the surface of many pathogens, including bacteria, viruses, fungi, and parasites.<sup>12</sup> This binding occurs through the carbohydrate-recognition domains of MBL. Following the binding of pathogens, MBL activates the lectin pathway of the complement system, resulting in a chain reaction of immune responses. This activation leads to the opsonization of pathogens, making them more visible and easier for phagocytic cells, such as macrophages and neutrophils, to ingest and destroy.<sup>13</sup> Furthermore, the complement activation can result in the formation of the membrane attack complex (MAC), which directly kills pathogens by creating pores in their cell membranes.<sup>14</sup> MBL also functions as an opsonin, a molecule that enhances phagocytosis by marking pathogens for destruction.

Phagocytes have receptors for MBL, enabling the uptake and clearance of the bound pathogens.<sup>14</sup> MBL has been associated with the modulation of immune responses in various innate immune-mediated conditions. These include autoimmune diseases such as systemic lupus erythematosus<sup>15,16</sup> and rheumatoid arthritis,<sup>17</sup> susceptibility to viral and bacterial infections,<sup>18</sup> and the regulation of inflammatory processes through the clearance of apoptotic and altered self-cells. Based on the significance of MBL, it has been hypothesized that individuals with lower levels of MBL may be more susceptible to developing sepsis (Details are summarized in Figure 1). Several investigations have been conducted worldwide to explore this relationship. The results of these studies have reported a strong correlation between lower levels of MBL and an increased risk of sepsis in adults and children.<sup>19-21</sup>

MBL is a protein encoded by the *MBL-2* gene, which is located on chromosome 10 at position 10q21.1. Several single-nucleotide polymorphisms (SNPs) have been identified in the promoter region of the *MBL-2* gene, and some of them have been shown to affect plasma levels of MBL. Two genetic variations at positions -550 (H > L, rs11003125) and -221 (Y > X, rs7096206) have been extensively studied as they are associated with alterations in plasma MBL levels.<sup>22-24</sup> The relationship between low MBL-producing genotypes of promoter polymorphisms and the development and clinical severity of sepsis has been studied, with some research indicating a connection.<sup>25-27</sup> However, other studies have not shown a similar link between MBL promoter polymorphisms and susceptibility to sepsis.<sup>28-31</sup> The inconsistent findings regarding the role of *MBL-2* promoter polymorphisms in sepsis may be due to smaller sample sizes in the studies, differences in the categorization of sepsis cases, and variations in the patients' ethnicity. Meta-analysis is a powerful technique that combines data from studies with similar designs, enhances the statistical power of the study, and draws a definitive conclusion.<sup>32</sup> In order to establish a definitive conclusion on the role of *MBL-2* promoter polymorphisms (H>L and Y>X) in sepsis susceptibility, we conducted a meta-analysis of all relevant published articles.

## MBL-2 Polymorphisms in Sepsis

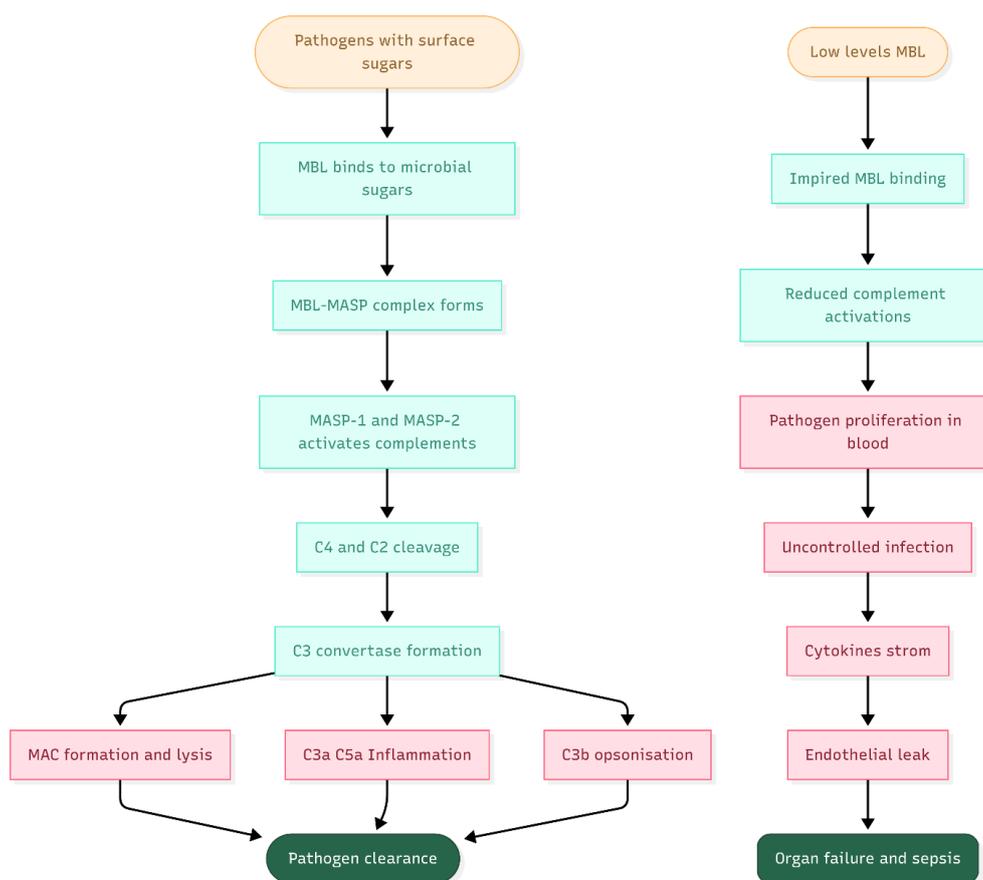


Figure 1. Schematic representation of the role of mannose-binding lectin in sepsis.

## MATERIALS AND METHODS

### The Search for Eligible Studies

The present meta-analysis incorporated articles from three databases, namely PubMed, Scopus, and ScienceDirect. To locate relevant articles, various keywords were employed, such as “Mannose-binding lectin,” “MBL,” “MBL-2,” “promoter polymorphism,” “H>L,” “Y>X,” “MBL-550,” “MBL-221,” “sepsis,” “septicemia,” and “septic shock.” Following a review of the titles and abstracts, eligible articles were selected, and their full texts were downloaded. Moreover, all eligible articles and their references were manually screened to identify additional relevant studies. Two authors independently conducted the database searches and assessed the titles and abstracts. The final database search was executed on 14th August 2025.

### Inclusion and Exclusion Criteria

The following inclusion and exclusion criteria

were established to maintain stringency in the meta-analysis before conducting the literature search: i) case-control study, ii) well-defined clinical characteristics of sepsis cases and controls, iii) investigation of genetic associations, including the *MBL-2* promoter polymorphisms (-550 H>L and -221 Y>X), iv) genotype or allele prevalence data for both cases and controls. The exclusion criteria for this analysis include: i) studies that focus on evaluation, case studies, animal investigations, or mechanistic aspects of the *MBL-2* polymorphisms, ii) reports that do not provide genotype or allele data, iii) data that were published in a conference or seminar as an abstract, and iv) studies that examine infection or bacteremia data rather than sepsis.

### Data Extraction

Data extraction was conducted independently by two authors for all eligible papers. The following data were extracted: first author's name, year of publication, genotype prevalence, allele frequency, genotyping

method, population details, and number of patients and controls. In the event of disagreements regarding paper selection or data extraction, both authors discussed and reached a consensus.

### Quality Assessment of the Included Reports

To assess the quality of studies, the Newcastle-Ottawa Scale (NOS) was employed.<sup>33</sup> All authors individually assessed the articles featured in this study and utilized the NOS score, which comprised three primary categories: i) subject selection rated at 4 stars, ii) comparison of subjects rated at 2 stars, and iii) clinical outcomes rated at 3 stars. A study with an overall score of 5 stars or higher typically exhibits good or fair quality.

### Statistical Analysis

In this study, the distribution of the *MBL-2* -550 H>L and -221 Y>X genotypes in healthy controls was analyzed for Hardy-Weinberg equilibrium (HWE) using a Microsoft Excel spreadsheet and the chi-square test. The power of each included report was assessed using the Gpower v3.1 software.<sup>34</sup> The Comprehensive Meta-Analysis v4 (CMA) software was employed for all meta-analysis-related statistics. Five genetic models were considered to compare the genotype and allele frequencies between sepsis and controls: i) allele contrast, ii) homozygous comparison, iii) heterozygous contrast, iv) dominant, and v) recessive model. The allele contrast model evaluates the overall impact of the variant allele, while the homozygous and heterozygous comparison models assess the effects of having two or one copy of the variant allele, respectively. The dominant model investigates whether carrying one or two copies of the variant allele similarly increases risk, whereas the recessive model focuses on the effect of having two copies compared to one or none. The potential for heterogeneity among the included studies was explored using Cochrane Q and I<sup>2</sup> analysis.<sup>35</sup> An I<sup>2</sup> value of less than 25% suggests an absence of heterogeneity, while a range of 25% to 50% indicates moderate heterogeneity. Values exceeding 50% demonstrate the presence of heterogeneity among the reports included.<sup>35</sup> For the meta-analysis, a random (heterogeneous) or fixed (homogeneous) model was employed based on the evaluation of heterogeneity.<sup>35</sup> The calculation of the combined odds ratio, 95% confidence interval, and probability values was undertaken, with a *p* value of less than 0.05 being deemed to demonstrate statistical significance.

Furthermore, a sensitivity analysis was conducted for all the tested genetic comparison models to evaluate the robustness of the meta-analysis. In this analysis, each study was sequentially excluded from the meta-analysis, and the results were compared to those of the parent analysis to determine the degree of deviation.<sup>36</sup> In addition, a trial sequential analysis was carried out to determine if there have been enough studies conducted thus far to establish a definitive conclusion about the association between the *MBL-2* promoter polymorphism and sepsis or if further investigations are necessary.<sup>37</sup>

## RESULTS

### Databases Search and Identification of Appropriate Reports

The widely recognized Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines, which were updated in 2020, were followed in the present meta-analysis.<sup>38,39</sup> The details of the search strategies employed to search for articles in three databases are provided in the Supplementary Table 1. A total of 821 articles were obtained using predefined keywords and a search strategy, with 53 articles from PubMed, 62 from Scopus, and 706 from Science Direct. Of these, 664 articles were excluded due to duplication in different databases and other reasons. Furthermore, articles with insufficient data or those that examined infections unrelated to sepsis were also excluded. Ultimately, seven studies<sup>25-31</sup> were deemed eligible for inclusion in the present study, and all of them contained data for the *MBL-2* Y>X polymorphism except for Huh et al,<sup>25</sup> which only had genotypic data for the *MBL-2* H>L polymorphism. In addition, Davis et al and Eisen et al provided both genotype and allele data for the *MBL-2* Y>X polymorphism. The findings of the eligible studies are depicted in the PRISMA flow chart (Figure 2). Additionally, a post-hoc power analysis was conducted for each study, with a lower effect size ( $w=0.15$ ) and an alpha error of 0.05, revealing that three studies, namely Huh et al,<sup>25</sup> Mills et al,<sup>30</sup> and Hellemann et al,<sup>29</sup> had significant power (over 80%) to detect any possible genetic association if it existed (Table 1). The quality assessment of individual eligible studies was performed by the NOS scale and is documented in Supplementary Table 2.

## MBL-2 Polymorphisms in Sepsis

**Table 1. Details of reports considered for the meta-analysis of the association of MBL-2 promoter polymorphisms (H>L and Y>X) with sepsis.**

<b>MBL-2 rs11003125 (H&gt;L)</b>															
Author and year of publication	Country (Ethnicity)	Sepsis type/ diagnosis criteria	Adults/Neonates	Severity Scores Assessment	Genotyping Methods	Healthy Control				HWE (p)	Sepsis Patients			Power of the study	
						(n)	HH	HL	LL		(n)	HH	HL	(%)	LL
Davis et al, 2010 <sup>28</sup>	USA (North America)	NR	Adults	NR	TaqMan SNP typing	53	19	25	9	0.87	24	12	9	3	14
Huh et al, 2009 <sup>25</sup>	Korea (East Asian)	American College of Chest Physicians/Society of Critical Care Medicine Consensus Conference in 1992	Adults	SOFA	PCR-sequencing	398	124	182	92	0.11	255	77	122	56	85.8
Eisen et al, 2006 <sup>27</sup>	Australia (Caucasian)	NR	Adults	SOFA	PCR-SSP	236	25	111	100	0.47	170	24	71	75	63.1
<b>MBL-2 rs7096206 (Y&gt;X)</b>															
First Author and year	Country	Sepsis type/ diagnosis criteria	Adults/Neonates	Severity scores Assessment	Genotyping methods	(n)	YY	YX	XX		(n)	YY	YX		XX
Mills et al, 2015 <sup>30</sup>	Europe continent (Mixed)	NR	Adults	NR, all ICU admitted	HRMA	477	140	246	91	0.35	590	173	299	118	98
Davis et al, 2010 <sup>28</sup>	USA (North America)	NR	Adults	NR	TaqMan SNP typing	53	4	16	33	0.31	28	3	7	18	14.6
Van Der Zwet et al, 2008 <sup>31</sup>	Netherlands (Caucasian)	NR	Neonates	All from NICU	TaqMan SNP typing	145	55	60	30	0.07	41	18	15	8	30.4
Helleman et al, 2007 <sup>29</sup>	Denmark (Caucasian)	NR	Adults	SOFA	PCR-SSP	533	150	265	118	0.96	185	48	99	38	89.3
Eisen et al, 2006 <sup>27</sup>	Australia (Caucasian)	NR	Adults	SOFA	PCR-SSP	236	144	81	11	0.92	170	97	63	10	63.1
Garred et al, 2003 <sup>26</sup>	Denmark (Caucasian)	American College of Chest Physicians/Society of Critical Care Medicine Consensus Conference in 1992	Adults	SOFA	PCR-SSP	250	125	73	52	<0.0001	197	117	41	39	68

HRMA: High resolution melting analysis; HWE: Hardy-Weinberg equilibrium; ICU: intensive care unit; MBL-2: Mannose-binding lectin-2; NICU: neonatal intensive care unit; NR: not reported; PCR-RFLP: polymerase chain reaction followed by restriction fragment length polymorphism; PCR-SSP: polymerase chain reaction-sequence-specific primer; SNP: single nucleotide polymorphism; SOFA: sequential organ failure assessment.

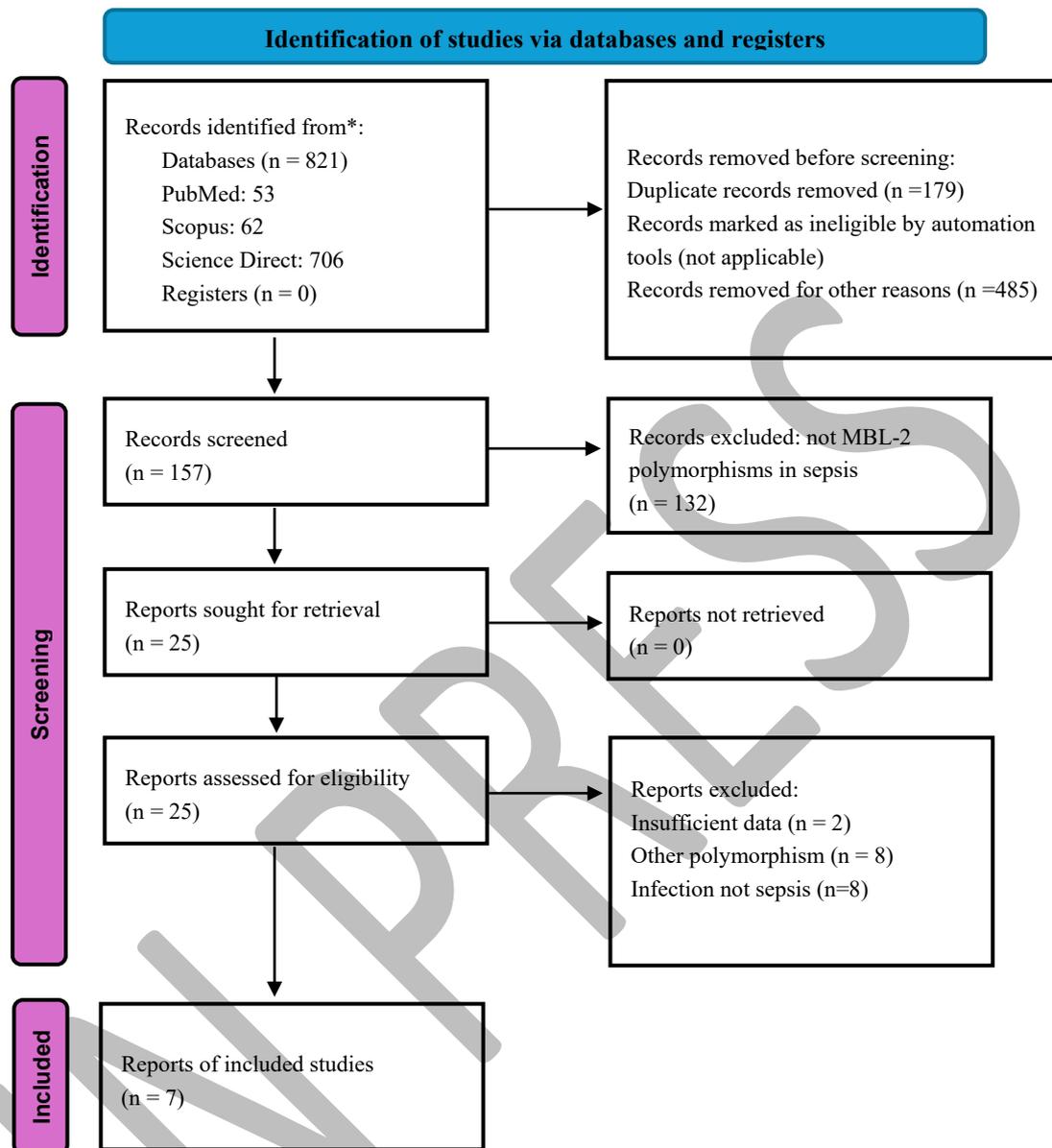


Figure 2. The inclusion process for the studies is depicted in the flowchart for the meta-analysis

### Publication Bias

The potential for publication bias in the included studies was assessed using Begg's funnel plot and Egger's regression analysis for all *MBL-2* H>L and Y>X polymorphisms genetic comparison models. As illustrated in Supplementary Figure 1, the funnel plots for *MBL-2* H>L showed no evidence of publication bias as they were symmetrical. Furthermore, Egger's regression also documented the absence of publication bias (Table 2). Similarly, for the other genetic variant, *MBL-2* Y>X, both the funnel plot and Egger's

regression analysis indicated the absence of publication bias (Supplementary Figure 2 and Table 2).

## MBL-2 Polymorphisms in Sepsis

**Table 2. Statistical analysis for publication bias and heterogeneity for the association of MBL2 promoter polymorphisms (H>L and Y>X) with sepsis.**

Comparison	Egger's Regression Analysis		Heterogeneity Analysis				Model Used for Meta-Analysis	Power (%)
	Intercept	95% Confidence interval	<i>p</i>	Q Value	<i>p</i> <sub>heterogeneity</sub>	I <sup>2</sup> (%)		
<b>MBL-2 rs11003125 (H&gt;L)</b>								
L vs H	-1.55	-3.70 to 0.58	0.06	1.08	0.58	0.00	Fixed	99.9
LL vs. HH	-1.23	-6.33 to 3.85	0.19	0.81	0.66	0.00	Fixed	83.4
HL vs. HH	-2.20	-13.07 to 8.67	0.23	2.52	0.28	20.86	Fixed	92.7
HL+LL vs. HH	-2.14	-8.03 to 3.75	0.13	2.18	0.33	8.24	Fixed	98.6
LL vs. HL+HH	-0.64	-9.85 to 8.55	0.53	0.48	0.78	0.00	Fixed	98.6
<b>MBL-2 rs7096206 (Y&gt;X)</b>								
X vs. Y	-0.43	-3.05 to 2.19	0.67	3.74	0.58	0.00	Fixed	100.0
XX vs YY	-0.24	-1.90 to 1.41	0.70	1.58	0.93	0.00	Fixed	99.9
YX vs. YY	-0.97	-4.15 to 2.20	0.44	6.38	0.27	21.73	Fixed	99.9
YX+XX vs. YY	-0.68	-3.79 to 2.43	0.57	5.34	0.37	6.40	Fixed	99.9
XX vs. YX+YY	0.13	-1.07 to 1.35	0.76	0.77	0.97	0.00	Fixed	99.9
<b>MBL-2 rs7096206 (Y&gt;X) Caucasian</b>								
X vs. Y	-0.43	-11.73 to 10.87	0.88	3.43	0.33	12.52	Fixed	100.0
XX vs YY	0.69	-4.58 to 5.97	0.62	1.20	0.75	0.00	Fixed	98.0
YX vs. YY	-2.33	-17.47 to 12.80	0.57	6.02	0.11	50.21	Fixed	99.7
YX+XX vs. YY	-0.90	-16.41 to 14.59	0.82	5.02	0.17	40.26	Fixed	99.9
XX vs. YX+YY	0.75	-1.61 to 3.12	0.30	0.48	0.92	0.00	Fixed	99.9

### Heterogeneity Test

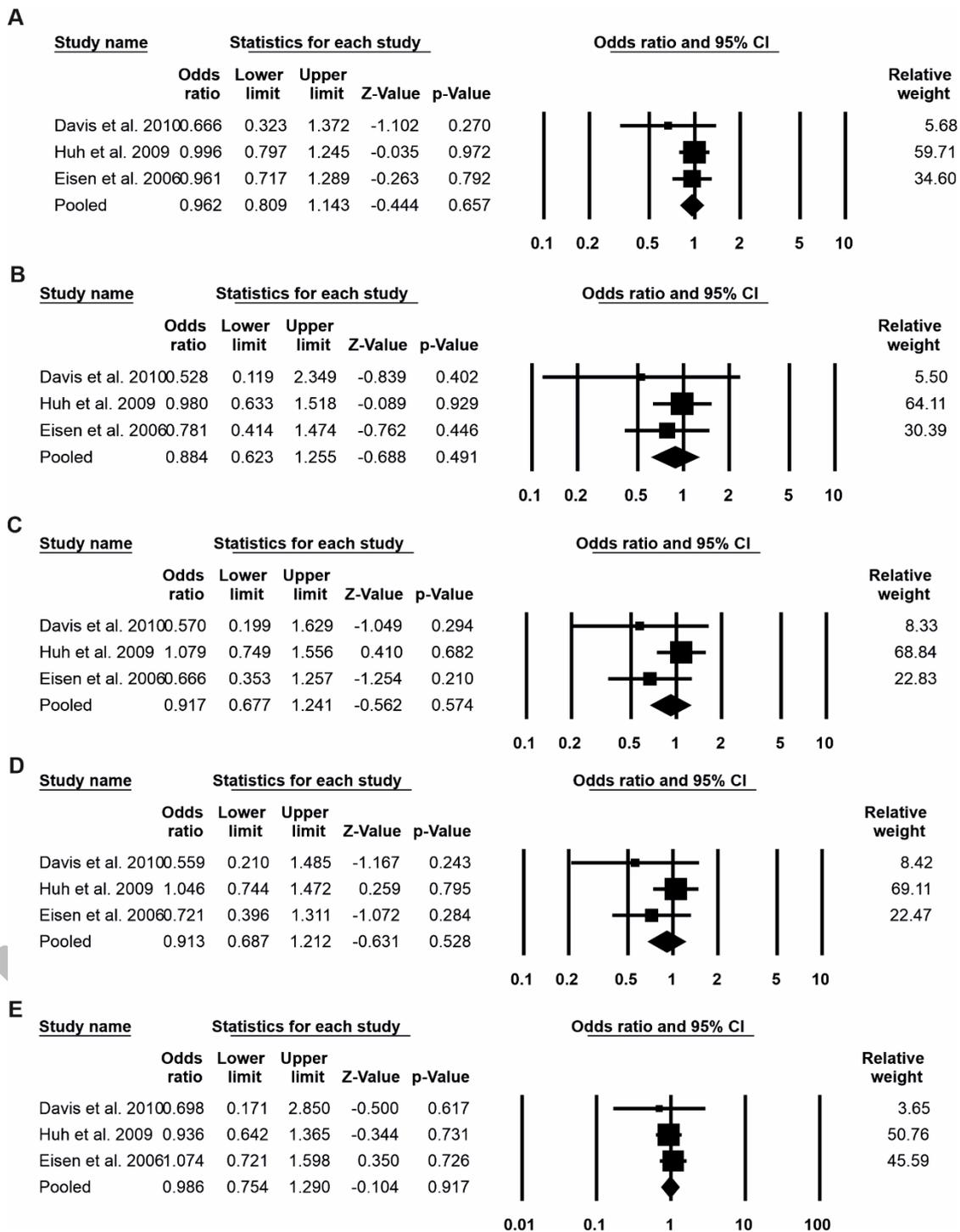
The analysis of heterogeneity among the studies was carried out using Cochran Q statistics and I<sup>2</sup> values. As shown in Table 2, there was significant homogeneity for all genetic comparison models for both the *MBL-2* H>L and Y>X polymorphisms. Consequently, a fixed effect model was chosen for the meta-analysis.

### MBL-2 H>L Polymorphism Is Not Associated with Sepsis

Three studies that met the eligibility criteria were included in the analysis to examine the role of the *MBL-2* H>L polymorphism in sepsis. These studies consisted of a total of 449 sepsis cases and 687 healthy controls. As illustrated in Figure 3, none of the genetic comparison models demonstrated a significant association between the *MBL-2* H>L promoter polymorphism and an increased risk of sepsis.

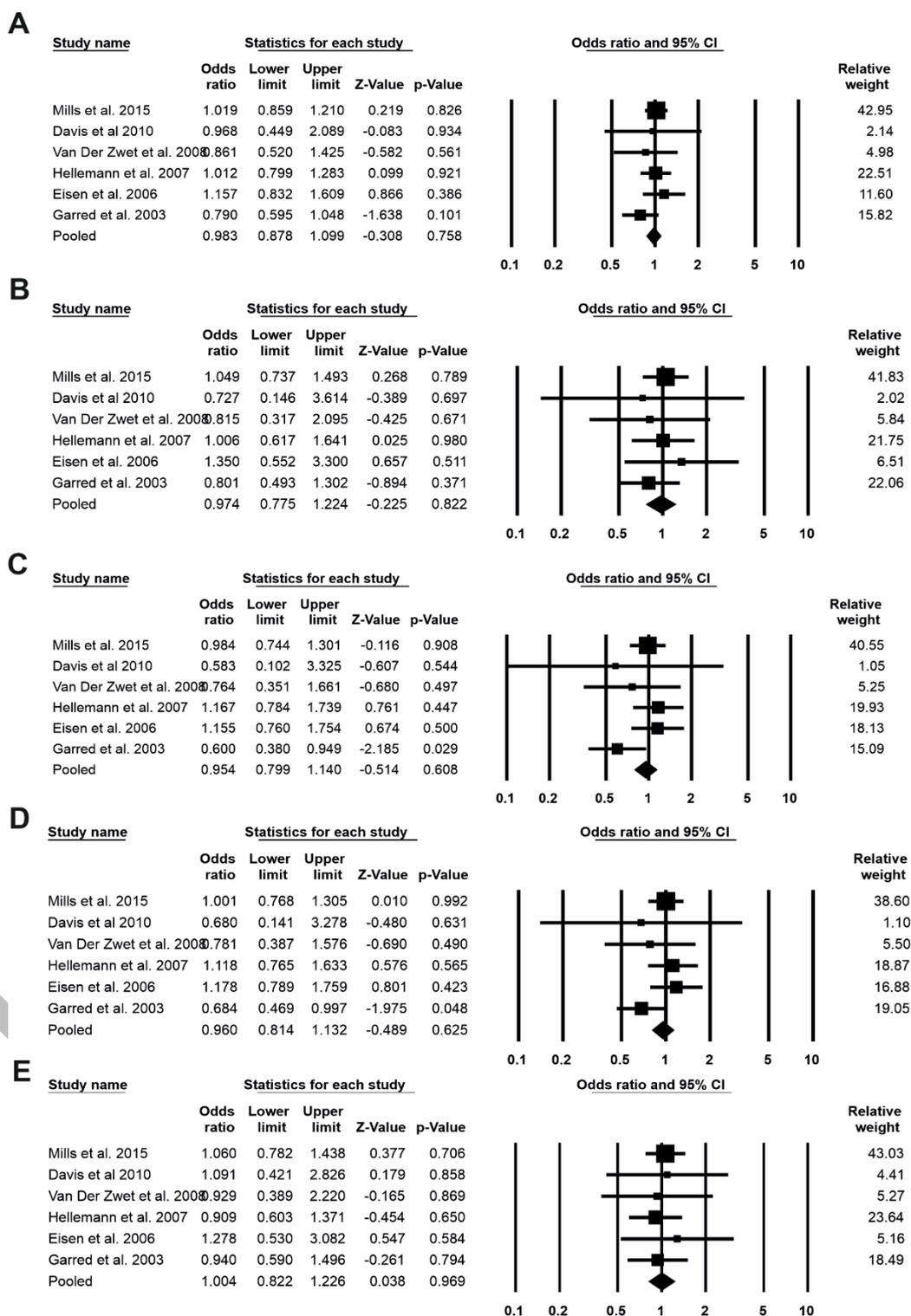
### MBL-2 Y>X Polymorphism Is Not Linked with Susceptibility to Sepsis

After assessing six case-control studies that were deemed eligible, we aimed to investigate the potential connection between *MBL-2* Y>X polymorphism and sepsis. A total of 1211 cases and 1694 controls were analyzed. As depicted in Figure 4, the distribution of alleles and genotypes was comparable between patients and healthy controls, which suggests that the *MBL-2* Y>X genetic variant does not play a role in the predisposition to sepsis.



**Figure 3.** Forest plots for the association of mannose-binding lectin (MBL)-2 H>L polymorphism with susceptibility to sepsis. The genetic association of MBL-2 promoter polymorphism (-550 H>L) with susceptibility to sepsis was performed by meta-analysis with the inclusion of three eligible reports. The genetic association was explored in five different genetic models comparison A. Allele, B. homozygous, C. heterozygous, D. dominant and E. recessive. All analysis was performed in CMA software.

## MBL-2 Polymorphisms in Sepsis



**Figure 4.** Forest plots for the association of mannose-binding lectin-2 Y>X polymorphism with susceptibility to sepsis. The genetic association of MBL-2 promoter polymorphism (-221 Y>X) with susceptibility to sepsis was performed by meta-analysis with the inclusion of six eligible reports. The genetic association was explored in five different genetic models comparison A) X vs. Y, B) XX vs. YY, C) YX vs YY, D) YX+XX vs. YY and E) XX vs. YX+YY. All analysis was performed in CMA software.

### Sensitivity Analysis

The sensitivity analysis in meta-analysis serves to illustrate the robustness of the analysis. By systematically eliminating one study at a time, conducting the meta-analysis, and comparing the deviation of the new analysis with the original analysis, the sensitivity analysis demonstrates the reliability of the meta-analysis. When there is no variation between the original and newer analyses, it provides evidence of the robustness of the meta-analysis.<sup>36</sup> We carried out a sensitivity analysis for the *MBL-2* promoter polymorphisms and found that the results showed minimal variations for both polymorphisms (H>L, as depicted in Supplementary Figure 3, and Y>X, as depicted in Supplementary Figure 4), which suggests that the meta-analysis findings for both polymorphisms are dependable.

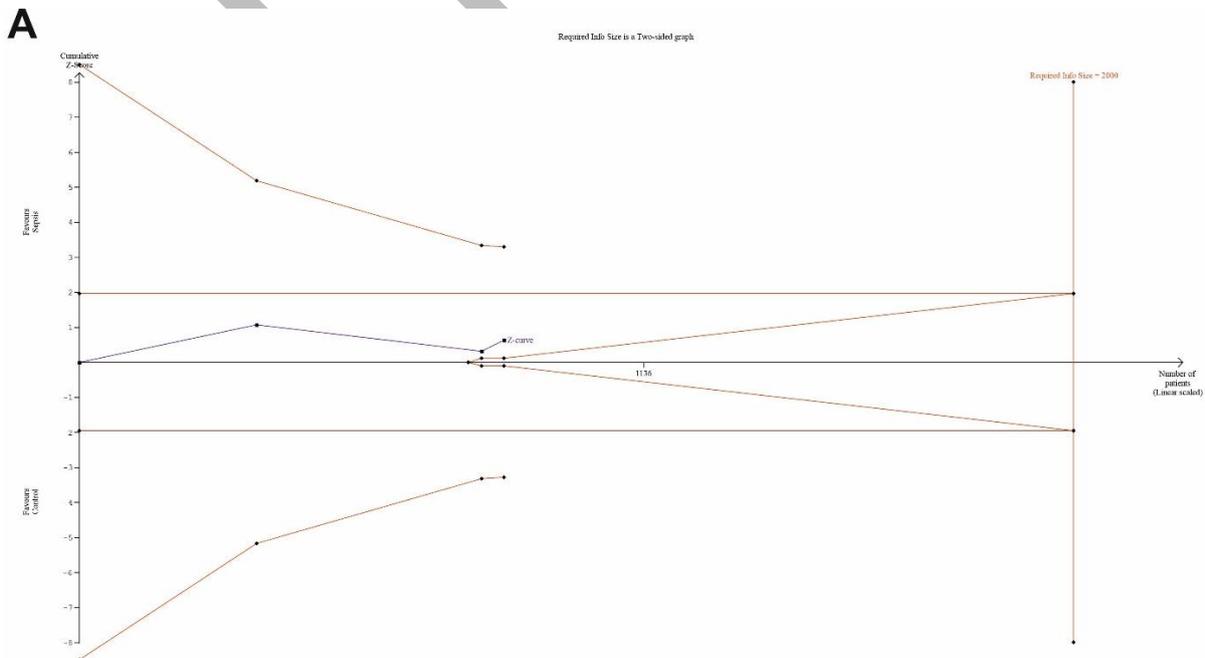
### Subgroup Analysis

The three reports concerning the *MBL2* H>L polymorphisms were derived from three distinct ethnic groups: North American, Caucasian, and East Asian. Consequently, due to the limited number of studies, a subgroup analysis could not be conducted. In contrast, among the six studies we considered for the *MBL* Y>X polymorphism, four were Caucasian, one was North American, and one was of mixed ethnicity. We conducted a subgroup analysis of the Caucasian population to investigate the association between the *MBL* Y>X polymorphism and sepsis. As shown in

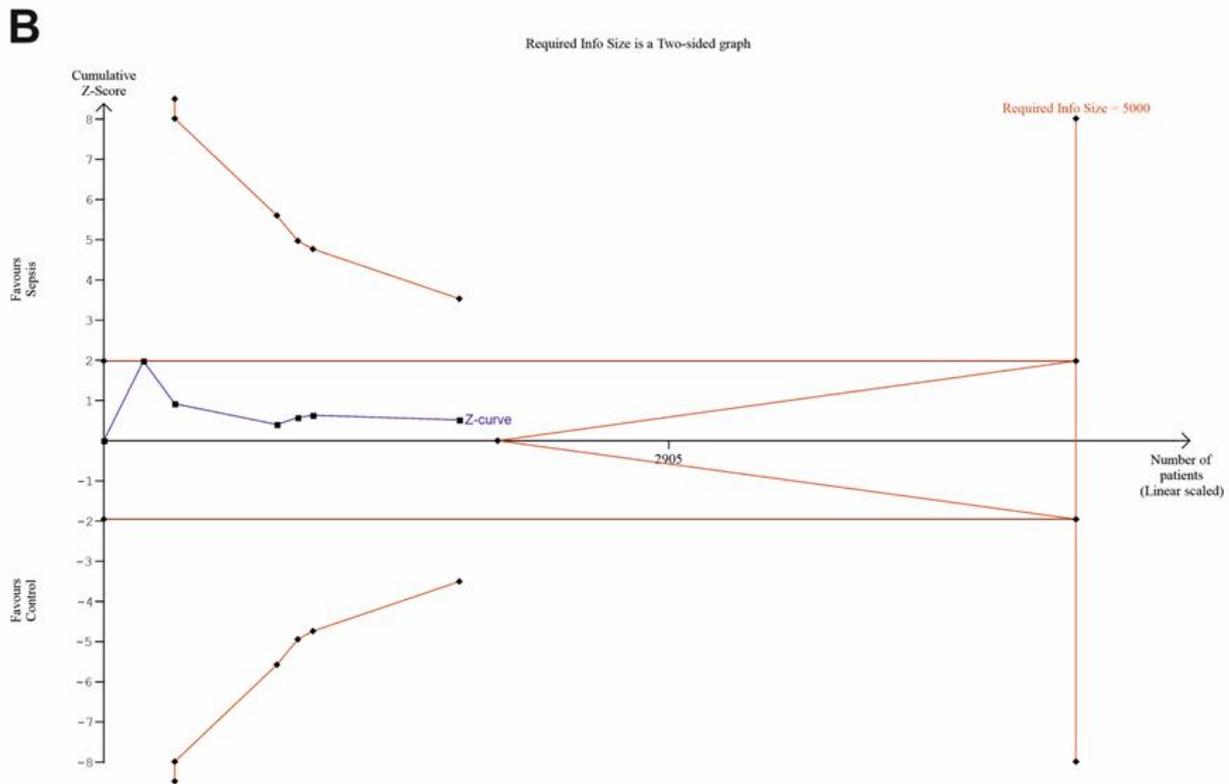
Supplementary Figure 5, no publication bias was observed for all genetic comparison models. No significant association was observed between the *MBL* Y>X polymorphism and susceptibility to sepsis in the Caucasian population (Supplementary Figure 6). Furthermore, the sensitivity analysis of the observations demonstrated robustness across all genetic comparison models (Supplementary Figure 7).

### Trial Sequential Analysis

The trial sequential analysis (TSA) methodologically arranges each study sequentially based on the year of publication, calculates the z statistics, and presents the results as a cumulative z-curve. The TSA plot features three important boundaries: alpha spending, futility, and required information size. When the cumulative z-curve intersects any of these boundaries, it signifies that enough studies have already been conducted, and further research is optional.<sup>37</sup> In the current investigation, we utilized genetic data from the dominant genetic model to prepare the TSA plot. Details of the parameter considered for the TSA are shown in Supplementary Table 3. As illustrated in Figure 5, the cumulative z curve did not surpass the alpha spending boundary, futility, or attain the necessary information size for both promoter polymorphisms of the *MBL-2* gene (H>L and Y>X). This suggests that additional genetic research is required to determine the role of *MBL-2* polymorphisms in sepsis susceptibility.



## MBL-2 Polymorphisms in Sepsis



**Figure 5.** Trial sequence analysis for the association of MBL promoter polymorphism (H>L and Y>X) with sepsis. TSA was performed to investigate the relationship between MBL-2 promoter variants and sepsis, with a focus on the H>L (A) and Y>X (B) polymorphisms. The crossing of the cumulative Z curve with the alpha spending boundary, the required information size line, or the futility boundary indicates that a sufficient number of investigations have been included in the meta-analysis, and no further studies are necessary. Conversely, if the cumulative Z curve does not intersect the alpha spending boundary, the futility boundary, or reach the required information size, it suggests that additional investigations are warranted. The results of the analysis revealed the need for further investigation to determine the association between these polymorphisms and sepsis.

### DISCUSSION

In this extensive analysis, seven studies were considered and analyzed to determine the relationship between *MBL-2* promoter polymorphisms (H>L and Y>X) and the predisposition to develop sepsis. The results of the present study indicate that there is no significant association between *MBL-2* promoter polymorphisms and susceptibility to sepsis. In addition, the sensitivity analysis supports the reliability of the meta-analysis, demonstrating minimal variation from the original meta-analysis when excluding one study each time and reperforming the analysis. Furthermore, the trial sequential analysis emphasizes the need for additional research to reach definitive conclusions about the role of *MBL-2* promoter polymorphisms in sepsis.

MBL is a vital component of the innate immune system, responsible for recognizing pathogen-associated molecular patterns and initiating the lectin pathway of complement activation.<sup>11</sup> The *MBL2* gene, which encodes for MBL, contains promoter polymorphisms at positions -550 (H>L) and -221 (Y>X) that influence the levels and functionality of MBL in serum.<sup>22-24</sup> The H>L polymorphism involves a C to G substitution, while the Y>X polymorphism involves a G to C substitution.<sup>40</sup> These genetic variations can lead to differential expression of MBL, which in turn can affect an individual's susceptibility to infections.<sup>24</sup> Studies have shown that individuals with low MBL levels, often associated with the L and X alleles, are at a higher risk of recurrent infections,<sup>41</sup> including respiratory tract infections<sup>42</sup> and systemic infections.<sup>43</sup> The role of *MBL*-

2 promoter polymorphisms has been extensively investigated, but the findings remain controversial.<sup>12,25,26,28-31</sup> Sepsis pathogenesis involves a complex interplay between pathogen load and the host's immune response,<sup>44</sup> where MBL plays a vital role in pathogen recognition and clearance. However, our meta-analysis and several other studies<sup>28-31</sup> have found no significant association between the -550 (H>L) and -221 (Y>X) polymorphisms and sepsis susceptibility. This suggests that while MBL levels influenced by these polymorphisms can affect the body's response to infections, they may not be a determining factor in sepsis development. Polymorphisms in the promoter region of *MBL2* not only regulate serum levels of MBL but may also influence the functional activity of other components within the lectin pathway. Given that MASP2 necessitates adequate MBL binding for complement activation<sup>45</sup> and FCN2 shares overlapping recognition functions,<sup>46</sup> a reduction in MBL availability due to promoter variants could impair both complement activation and cooperative pathogen clearance. This suggests that the impact of *MBL2* promoter polymorphisms on sepsis susceptibility may extend beyond MBL alone, affecting the overall efficiency of the lectin pathway.

Three meta-analyses<sup>47-49</sup> have been performed thus far to evaluate the link between *MBL-2* promoter polymorphisms and susceptibility to sepsis. Two of these investigations were conducted within the context of neonatal sepsis, encompassing 7 and 17 studies, respectively.<sup>47,48</sup> Both reports examined the association between the MBL structural variant and susceptibility to sepsis development. Luo et al<sup>47</sup> incorporating 338 cases and 1066 controls, demonstrated that the structural variant is associated with sepsis susceptibility. In contrast, Jakovljevic et al<sup>48</sup> did not document an association of the structural variant in culture-proven sepsis (n=6) or clinical plus culture-proven sepsis (n=11), considering 972 cases and 4622 controls, or 3337 cases and 5334 controls, respectively. In addition, a third meta-analysis was carried out with pediatric and adult sepsis patients, and it also failed to demonstrate an association,<sup>49</sup> similar to our current study. Notably, the number of studies considered in the meta-analysis exceeded those in the present report: specifically, 16 reports were included for Y>X, and seven studies were considered for H>L.<sup>49</sup> The report employed three genetic comparison models (allele contrast, dominant, and recessive) for their analysis, and the majority of the studies included in their meta-analysis had

a quality score of less than six.<sup>49</sup> The maintenance of rigorous inclusion and exclusion criteria is crucial in conducting a meta-analysis. Our analysis was limited to reports that employed appropriate definitions of sepsis patients. Moreover, earlier studies were examined for infection data, but it is important to note that infections and sepsis are clinically distinct. Furthermore, subgroup analysis within the Caucasian population indicated the absence of MBL promoter variants associated with susceptibility to sepsis. However, we were unable to explore other populations due to the limited number of studies available.

#### STATEMENT OF ETHICS

This study is a systematic review and meta-analysis of previously published studies and does not involve any new studies with human participants or animals. Therefore, ethical approval and informed consent were not required, in accordance with the institutional and national guidelines.

#### FUNDING

No specific funding has been received.

#### CONFLICT OF INTEREST

The authors declare no conflicts of interest.

#### ACKNOWLEDGMENTS

We wish to thank all the faculty members of the department for their constant support and guidance.

#### DATA AVAILABILITY

Upon reasonable request to the corresponding author

#### AI ASSISTANCE DISCLOSURE

No artificial intelligence (AI) tools were used in preparing this manuscript.

#### REFERENCES

1. Jarczak D, Kluge S, Nierhaus A. Sepsis-pathophysiology and therapeutic concepts. *Frontiers in medicine*. 2021;8:628302.

## MBL-2 Polymorphisms in Sepsis

- Rudd KE, Johnson SC, Agesa KM, Shackelford KA, Tsoi D, Kievlan DR, et al. Global, regional, and national sepsis incidence and mortality, 1990–2017: analysis for the Global Burden of Disease Study. *The Lancet*. 2020;395(10219):200-11.
- Markwart R, Saito H, Harder T, Tomczyk S, Cassini A, Fleischmann-Struzek C, et al. Epidemiology and burden of sepsis acquired in hospitals and intensive care units: a systematic review and meta-analysis. *Intensive Care Med*. 2020;46(8):1536-51.
- Arefian H, Heublein S, Scherag A, Brunkhorst FM, Younis MZ, Moerer O, et al. Hospital-related cost of sepsis: a systematic review. *Journal of Infection*. 2017;74(2):107-17.
- Honda TJ, Kazemiparkouhi F, Henry TD, Suh HH. Long-term PM2.5 exposure and sepsis mortality in a US medicare cohort. *BMC Public Health*. 2022;22(1):1214.
- Lu H, Wen D, Wang X, Gan L, Du J, Sun J, et al. Host genetic variants in sepsis risk: a field synopsis and meta-analysis. *Critical Care*. 2019;23(1):26.
- Pradeu T, Thomma BPHJ, Girardin SE, Lemaitre B. The conceptual foundations of innate immunity: Taking stock 30 years later. *Immunity*. 2024;57(4):613-31.
- Nie J, Zhou L, Tian W, Liu X, Yang L, Yang X, et al. Deep insight into cytokine storm: from pathogenesis to treatment. *Signal Transduction and Targeted Therapy*. 2025;10(1):112.
- Hotchkiss RS, Monneret G, Payen D. Sepsis-induced immunosuppression: from cellular dysfunctions to immunotherapy. *Nature Reviews Immunology*. 2013;13(12):862-74.
- Wiersinga WJ, van der Poll T. Immunopathophysiology of human sepsis. *eBioMedicine*. 2022;86.
- Takahashi K, Ezekowitz RA. The role of the mannose-binding lectin in innate immunity. *Clin Infect Dis*. 2005;41 Suppl 7:S440-4.
- Eisen DP, Minchinton RM. Impact of mannose-binding lectin on susceptibility to infectious diseases. *Clin Infect Dis*. 2003;37(11):1496-505.
- Ip WE, Takahashi K, Moore KJ, Stuart LM, Ezekowitz RAB. Mannose-binding lectin enhances Toll-like receptors 2 and 6 signaling from the phagosome. *The Journal of experimental medicine*. 2008;205(1):169-81.
- Tegla CA, Cudrici C, Patel S, Trippe R, Rus V, Niculescu F, et al. Membrane attack by complement: the assembly and biology of terminal complement complexes. *Immunologic Research*. 2011;51(1):45-60.
- Panda AK, Parida JR, Tripathy R, Pattanaik SS, Ravindran B, Das BK. Low producer MBL genotypes are associated with susceptibility to systemic lupus erythematosus in Odisha, India. *Hum Immunol*. 2013;74(1):114-9.
- Panda AK, Parida JR, Tripathy R, Pattanaik SS, Ravindran B, Das BK. Mannose binding lectin: a biomarker of systemic lupus erythematosus disease activity. *Arthritis Res Ther*. 2012;14(5):R218.
- Graudal NA, Homann C, Madsen HO, Svejgaard A, Jurik AG, Graudal HK, et al. Mannan binding lectin in rheumatoid arthritis. A longitudinal study. *J Rheumatol*. 1998;25(4):629-35.
- Gupta K, Gupta RK, Hajela K. Disease associations of mannose-binding lectin & potential of replacement therapy. *Indian J Med Res*. 2008;127(5):431-40.
- Jacobson S, Larsson P, Åberg AM, Johansson G, Winsö O, Söderberg S. Levels of mannose-binding lectin (MBL) associates with sepsis-related in-hospital mortality in women. *J Inflamm (Lond)*. 2020;17:28.
- Frakking FN, Brouwer N, van Eijkelenburg NK, Merkus MP, Kuijpers TW, Offringa M, et al. Low mannose-binding lectin (MBL) levels in neonates with pneumonia and sepsis. *Clin Exp Immunol*. 2007;150(2):255-62.
- Gordon AC, Waheed U, Hansen TK, Hitman GA, Garrard CS, Turner MW, et al. Mannose-binding lectin polymorphisms in severe sepsis: relationship to levels, incidence, and outcome. *Shock*. 2006;25(1):88-93.
- Mao Y, Wei H, Gong Y, Peng L, Chen Y. Association of MBL2 gene polymorphisms and MBL levels with dilated cardiomyopathy in a Chinese Han population. *BMC Medical Genomics*. 2024;17(1):11.
- Tsakanova G, Stepanyan A, Steffensen R, Soghoyan A, Jensenius JC, Arakelyan A. Pattern recognition molecules of lectin complement pathway in ischemic stroke. *Pharmacogenomics and personalized medicine*. 2021:1347-68.
- Das BK, Panda AK. MBL-2 polymorphisms (codon 54 and Y-221X) and low MBL levels are associated with susceptibility to multi organ dysfunction in *P. falciparum* malaria in Odisha, India. *Front Microbiol*. 2015;6:778.
- Huh JW, Song K, Yum J-S, Hong S-B, Lim C-M, Koh Y. Association of mannose-binding lectin-2 genotype and serum levels with prognosis of sepsis. *Critical Care*. 2009;13:1-9.
- Garred P, J. Strøm J, Quist L, Taaning E, Madsen HO. Association of mannose-binding lectin polymorphisms with sepsis and fatal outcome, in patients with systemic inflammatory response syndrome. *The Journal of infectious diseases*. 2003;188(9):1394-403.
- Eisen DP, Dean MM, Thomas P, Marshall P, Gerns N,

- Heatley S, et al. Low mannose-binding lectin function is associated with sepsis in adult patients. *FEMS Immunology & Medical Microbiology*. 2006;48(2):274-82.
28. Davis SM, Clark EA, Nelson LT, Silver RM. The association of innate immune response gene polymorphisms and puerperal group A streptococcal sepsis. *American Journal of Obstetrics and Gynecology*. 2010;202(3):308. e1-8.
  29. Hellemann D, Larsson A, Madsen HO, Bonde J, Jarlov JO, Wiis J, et al. Heterozygosity of mannose-binding lectin (MBL2) genotypes predicts advantage (heterosis) in relation to fatal outcome in intensive care patients. *Human molecular genetics*. 2007;16(24):3071-80.
  30. Mills TC, Chapman S, Hutton P, Gordon AC, Bion J, Chiche J-D, et al. Variants in the mannose-binding lectin gene MBL2 do not associate with sepsis susceptibility or survival in a large European cohort. *Clinical Infectious Diseases*. 2015;61(5):695-703.
  31. Van Der Zwet W, Catsburg A, Van Elburg R, Savelkoul P, Vandenbroucke-Grauls C. Mannose-binding lectin (MBL) genotype in relation to risk of nosocomial infection in pre-term neonates in the neonatal intensive care unit. *Clinical Microbiology and Infection*. 2008;14(2):130-5.
  32. Borenstein M, Hedges LV, Higgins JP, Rothstein HR. *Introduction to meta-analysis*: John Wiley & Sons; 2021.
  33. Wells GA, Shea B, O'Connell D, Peterson J, Welch V, Losos M, et al. The Newcastle-Ottawa Scale (NOS) for assessing the quality of nonrandomised studies in meta-analyses. 2000.
  34. Erdfelder E, Faul F, Buchner A. GPOWER: A general power analysis program. *Behavior research methods, instruments, & computers*. 1996;28:1-11.
  35. Higgins JP, Thompson SG. Quantifying heterogeneity in a meta-analysis. *Statistics in medicine*. 2002;21(11):1539-58.
  36. Copas J, Shi JQ. Meta-analysis, funnel plots and sensitivity analysis. *Biostatistics*. 2000;1(3):247-62.
  37. Wetterslev J, Thorlund K, Brok J, Gluud C. Trial sequential analysis may establish when firm evidence is reached in cumulative meta-analysis. *Journal of clinical epidemiology*. 2008;61(1):64-75.
  38. Page MJ, McKenzie JE, Bossuyt PM, Boutron I, Hoffmann TC, Mulrow CD, et al. The PRISMA 2020 statement: an updated guideline for reporting systematic reviews. *bmj*. 2021;372.
  39. Panda AK. Maintaining ethics, Integrity, and accountability: Best practices for reporting a meta-analysis. *Account Res*. 2024:1-3.
  40. Mahto H, Pati A, Sahu SK, Sharma HP, Padhi A, Panda AK. Association of MBL-2 gene polymorphisms with systemic lupus erythematosus: an updated meta-analysis and trial sequential analysis. *Lupus*. 2020;29(10):1227-37.
  41. Mullighan CG, Marshall SE, Welsh KI. Mannose binding lectin polymorphisms are associated with early age of disease onset and autoimmunity in common variable immunodeficiency. *Scand J Immunol*. 2000;51(2):111-22.
  42. Rantala A, Lajunen T, Juvonen R, Bloigu A, Silvennoinen-Kassinen S, Peitso A, et al. Mannose-binding lectin concentrations, MBL2 polymorphisms, and susceptibility to respiratory tract infections in young men. *Journal of Infectious Diseases*. 2008;198(8):1247-53.
  43. Turner MW. Mannose-binding lectin (MBL) in health and disease. *Immunobiology*. 1998;199(2):327-39.
  44. van Engelen TSR, Joost Wiersinga W, van der Poll T. Pathogenesis of Sepsis. In: Wiersinga WJ, Seymour CW, editors. *Handbook of Sepsis*. Cham: Springer International Publishing; 2018. p. 31-43.
  45. Beltrame MH, Boldt AB, Catarino SJ, Mendes HC, Boschmann SE, Goeldner I, et al. MBL-associated serine proteases (MASPs) and infectious diseases. *Mol Immunol*. 2015;67(1):85-100.
  46. Meijvis SCA, Herpers BL, Endeman H, de Jong B, van Hanne E, van Velzen-Blad H, et al. Mannose-binding lectin (MBL2) and ficolin-2 (FCN2) polymorphisms in patients on peritoneal dialysis with staphylococcal peritonitis. *Nephrology Dialysis Transplantation*. 2010;26(3):1042-5.
  47. Luo J, Xu F, Lu GJ, Lin HC, Feng ZC. Low mannose-binding lectin (MBL) levels and MBL genetic polymorphisms associated with the risk of neonatal sepsis: An updated meta-analysis. *Early Hum Dev*. 2014;90(10):557-64.
  48. Sljivancanin Jakovljevic T, Martic J, Jacimovic J, Nikolic N, Milasin J, Mitrović TL. Association between innate immunity gene polymorphisms and neonatal sepsis development: a systematic review and meta-analysis. *World J Pediatr*. 2022;18(10):654-70.
  49. Zhang AQ, Yue CL, Pan W, Gao JW, Zeng L, Gu W, et al. Mannose-binding lectin polymorphisms and the risk of sepsis: evidence from a meta-analysis. *Epidemiol Infect*. 2014;142(10):2195-206.
  50. Westwood ME, Whiting PF, Kleijnen J. How does study quality affect the results of a diagnostic meta-analysis? *BMC Med Res Methodol*. 2005;5:20.